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Identification of the Water Quality Factors Which Prevent Fingernail Clams from Recolonizing the Illinois River, Phase III

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IDENTIFICATION OF THE WATER QUALITY FACTORS WHICH PREVENT FINGERNAIL CLAMS FROM RECOLONIZING THE ILLINOIS RIVER

PHASE III

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TECHNICAL COMPLETION REPORT

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Water Resources Center
University of Illinois at Urbana-Champaign
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TABLE OF CONTENTS

TABLE OF CONTENTS (continued)

LIST OF FIGURES

LIST OF TABLES

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ABSTRACT

The purpose of this research was to determine why fingernail clams have been unable to recolonize a 100-mile reach of the Illinois River where they were abundant prior to a die-off in the 1950's. Fingernail clams are major links in food chains leading from detritus and algae to higher level consumers valued by man, such as fish and water fowl. Three suspected toxicants and sediments from the reach where the die-off occurred were tested on intact fingernail clams (*Musculium transversum*) and gill preparations isolated from the clams.

Concentrations of fluoride, lead and cadmium which caused a 50% reduction in the rate of beating of cilia on isolated clam gills, after 10 minutes of exposure (10-minute EC50), were 0.75, 0.02 and 0.06 mg/l, respectively. Mixtures of cadmium and fluoride were sightly more toxic to clam gills than predicted from results of bioassays with single toxicants. A fluoride concentration of 2.82 mg/l killed intact figernail clams after eight weeks of exposure, while mortality in lesser concentrations and one higher concentration did not differ significantly from controls maintained in well water to which no fluoride had been added. Hence, the sublethal response exhibited by the gills is at least four times more sensitive than the lethal response. Maximum fluoride concentrations reported by the U.S. Geological Survey at two stations in the Illinois River ranged from .6 to .8 mg/l between 1979 and 1981, considerably below the concentrations which affected growth and survival of intact clams during 8-week exposures in our laboratory, but slightly above the level which affected isolated clam gills. A lead bioassay using intact clams was completed, but the results were ambiguous because concentration ranges in separate test chambers overlapped. In addition, insoluble lead precipitates accumulated in the test chambers, and the relative toxicity to clams of the soluble versus the insoluble lead was not determined. Until additional bioassays are completed, it is impossible to determine whether the maximum total lead concentrations of 0.40 mg/l which occurred in the Illinois River between 1979 and 1981 could have contributed to the failure of fingernail clams to recolonize the river.

Fingernail clams exposed to sediments from lakes along the Illinois River suffered greater mortality after six weeks of exposure than clams exposed to sediment from the Mississippi River, although the differences were not statistically significant. The same sediments tested on clam gills produced statistically significant changes in ciliary beating rate and particle transport rates on the gills. Sediments from the upstream lakes cause a greater depression in the particle transport rate and ciliary beating rates than sediments from downstream lakes. In addition, sediments from the lake furthest upstream caused a drastic change from the normal metachronal beating pattern to an atypical synchronous pattern.

The results with the gill assay suggest that sediments in the Illinois River contain unidentified toxic factors and that sediments in the upper river, closer to the metropolitan areas of Joliet and Chicago, are more toxic than sediments further downstream. These results should be confirmed by additional tests with intact clams, including field tests with caged organisms. Parallel chemical analyses and bioassays of extracts from the sediments should be performed to identify the toxic components.

Sparks, Richard E., Michael J. Sandusky and Anthony A. Paparo

IDENTIFICATION OF THE WATER QUALITY FACTORS WHICH PREVENT FINGERNAIL CLAMS FROM RECOLONIZING THE ILLINOIS RIVER, PHASE III
Technical Completion Report to the Bureau of Reclamation, Office of Water Policy, April 1983

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INTRODUCTION AND BACKGROUND ON THE ILLINOIS RIVER

COMMERCIAL FISH

In view of the fact that over four billion dollars has been spent since the mid-1960's on waste treatment by municipalities in the Illinois River drainage basin (Briceland, 1976), it is surprising that the fish catch from the Illinois River has shown a steady decline since 1950, while the catch on the Mississippi has remained relatively constant (Figure 1). Although the number of full-time commercial fishermen on both rivers has declined since 1950, the number of

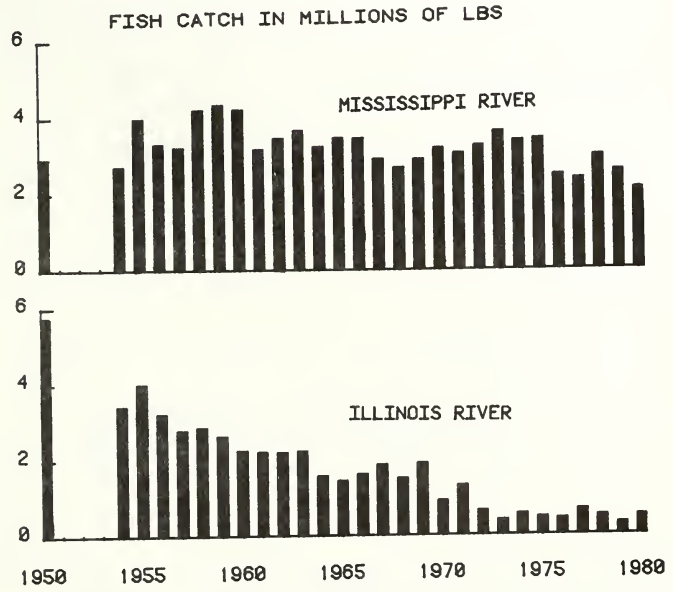

Figure 1. Commercial fish catch from the Illinois River and from the Mississippi River bordering Illinois. Source: Illinois Department of Conservation.

full-time fishermen on the Mississippi bordering Illinois has stabilized since 1964, while the number of full-time fishermen on the Illinois has continued to decline (Figure 2). While the recent declines in the number of commercial fishermen are dramatic, they are even more startling in comparison tc 1908, the peak year for the Illinois River commercial fishery. In 1908, the value of the catch exceeded that of any other river in America (excluding rivers with anadromous fishes) and

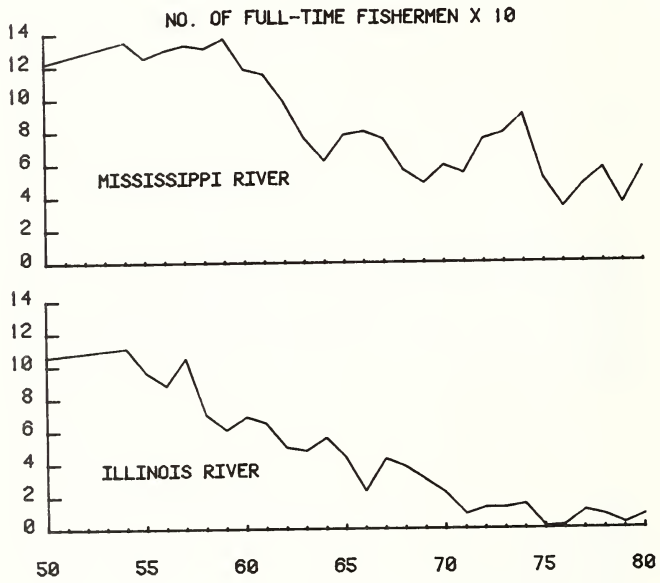

Figure 2. Number of full-time commercial fishermen on the Illinois River and on the Mississippi River bordering Illinois. Source: Illinois Department of Conservation.

over 2,000 commercial fishermen found employment on the river (Department of Commerce and Labor, 1911). The harvest was 10 percent of the total U.S. freshwater harvest (Department of Commerce and Labor, 1911). In 1976, only two full-time commercial fishermen worked on the Illinois River, and the 1973 harvest was only 0.32 percent of the total U.S. harvest of freshwater fish (Department of Commerce, 1980 and 1976).

Economic factors alone do not explain the decline in fishing effort and catch on the Illinois River. The real value of Illinois River fish has remained relatively constant since the 1950's. In fact, when the wholesale prices are expressed in constant 1981 cents, current prices for drum and buffalo are about the same as in 1894, carp prices have decreased, and channel catfish prices have substantially increased (Sparks, in press).

The annual per-acre yield of fish from the Upper Mississippi River has actually increased slightly (Lubinski, Wallendorf, and Reese, 1981), while yield from the Illinois River is only a tenth of what it was in the 1950's (Lubinski, Wallendorf, and Reese, 1981) and only a fiftieth of what it was in 1908 (Richardson, 1921):

Upper Mississippi	lbs/acre	Illinois	lbs/acre
		1908	178.4
1953-62	26.3	1950-59	38.1
1973-77	29.2	1973-80	3.7

The recent decline in the commercial fishery of the Illinois River appears to be related to a decline in the food supply for the fish. Starrett (1972) studied the food habits of carp, a commercially important species, in the Illinois River in the 1960's and found that in the lower Illinois River fingernail clams comprised 50% of the volume of food items in carp stomachs, whereas in the middle and upper section of the river only one fingernail clam was found in all the stomachs examined. In 1963, the condition factor of carp was considerably better in the lower Illinois River than in the middle and upper sections (Figure 3).

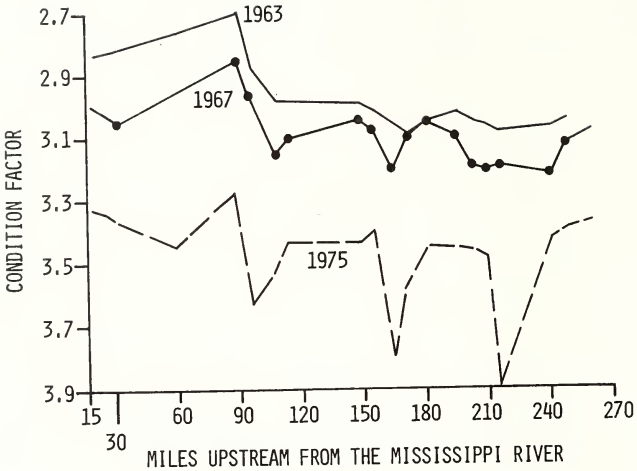

Figure 3. Condition factor of carp (body length divided by body depth) in the Illinois River in 1963, 1967, and 1975. Source: Illinois Natural History Survey.

Channel catfish were also more abundant in the lower Illinois River (Starrett, 1972). The condition factor of carp in 1967 showed the same pattern as in 1963, but an overall decline had occurred, with more pronounced declines at river miles 95-105, 160-170, and 200-240. By 1975, the overall condition of carp had declined further, and localized declines had become more severe. If the 1975 data for condition factor of carp are plotted against the biomass of bottom fauna, a high degree of correlation is evident (Figure 4).

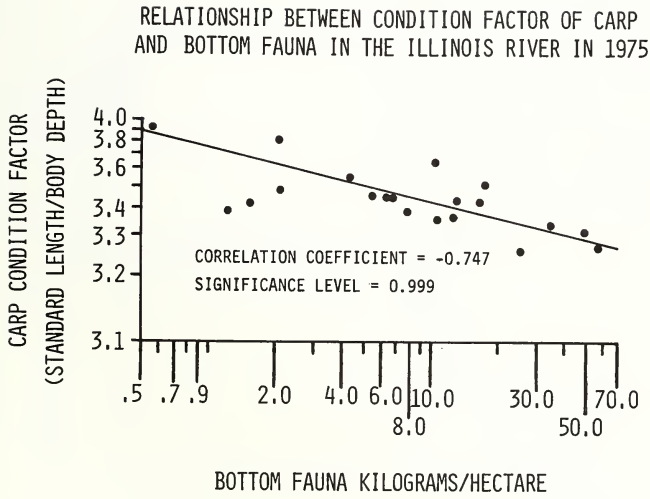

Figure 4. Relationship between condition factor (body length divided by body depth: high numbers indicate a thin fish, of little market value) of carp and biomass of benthos in Illinois River in 1975. Source: Illinois Natural History Survey.

Prior to the 1950's, the greatest harvest of commercial fish generally occurred along the middle section of the Illinois River near Havana, in areas where food organisms, such as fingernail clams were most abundant (Richardson, 1921). Paloumpis and Starrett (1960) documented a die-off of fish food organisms in the middle section of the Illinois River in the mid-1950's (Figure 5), and more recent surveys

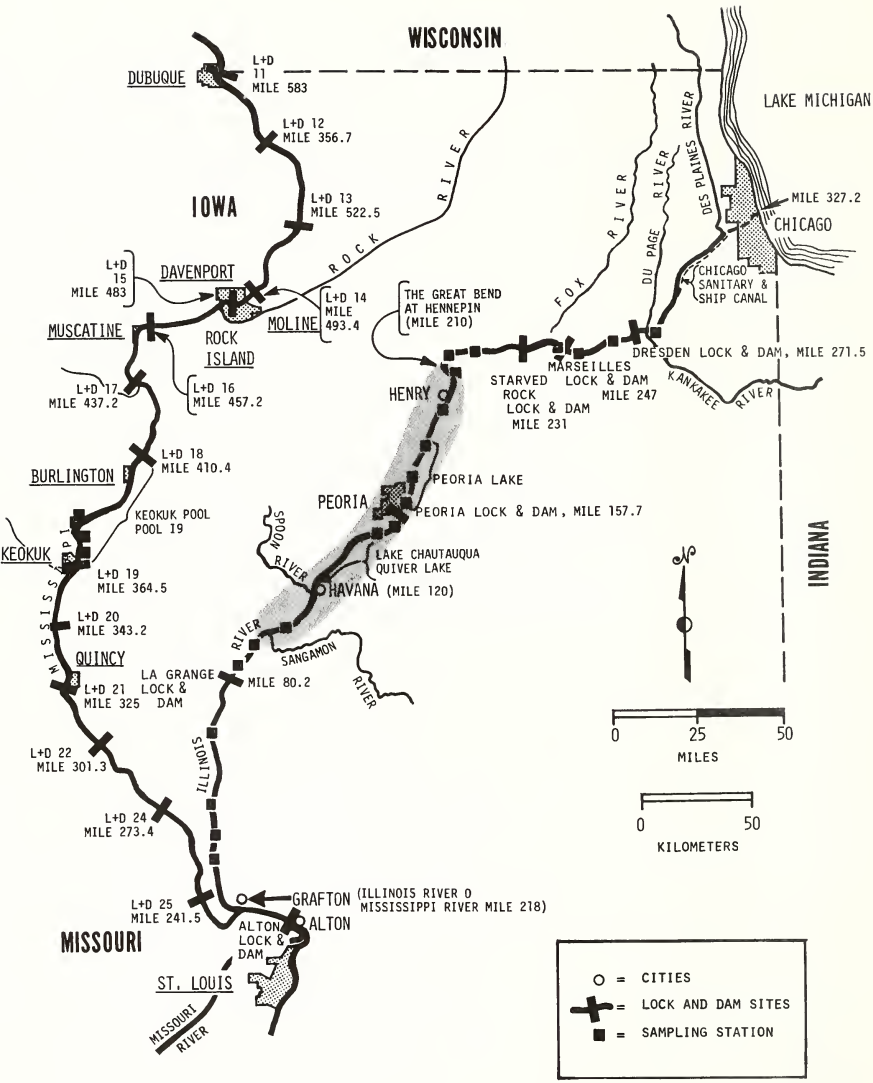

Figure 5. Map of the Illinois River and the Upper Mississippi River, bordering Illinois. Benthos died out in the shaded area.

show that the clams have failed to recolonize areas where they were formerly abundant (Anderson, 1977; Anderson, Sparks, and Paparo, 1978). The decline in condition factor and growth of commercial species of fishes thus is clearly related to the decline in benthos of the Illinois River and its connecting lakes.

DIVING DUCKS

Diving ducks also feed on benthic organisms, and use of the Illinois River by lesser scaup ducks plummeted at about the same time as the die-off of fingernail clams and snails (Figure 6). The duck

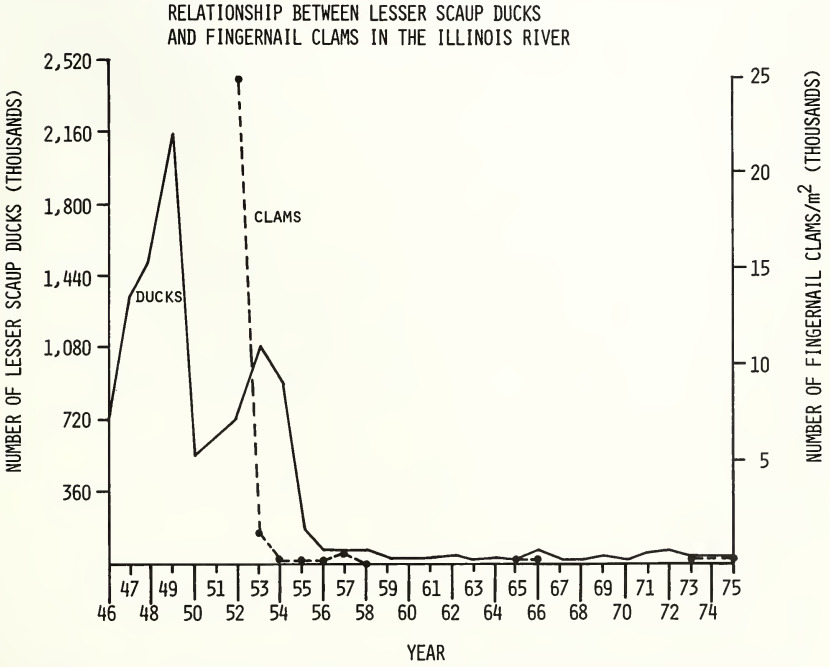

Figure 6. Relationship between fingernail clams and use of the Illinois River by diving ducks. Source: Illinois Natural History Survey. Duck population data furnished by F.C. Bellrose, Waterfowl Specialist.

population data in Figure 6 is for the entire Illinois River valley, whereas the fingernail clam data is for Quiver Lake only. If the die-off of benthic organisms did not occur simultaneously, but progressively in the affected 100-mile reach, ducks may have continued using the lower part of the reach after the populations in Quiver Lake had already died out. If the number of lesser scaup ducks utilizing Quiver Lake is superimposed on the clam populations in the lake, both populations show a simultaneous decline in the spring of 1953 (Personal communication, Dr. Frank C. Bellrose, Waterfowl Specialist, Wildlife Section, Illinois Natural History Survey). Use of the Mississippi River bordering Illinois increased, as the ducks apparently shifted their migration route away from the Illinois and over to the Mississippi, where dense populations of fingernail clams occurred in places such as Pool 19 (Personal communication, Dr. Frank C. Bellrose, Waterfowl Specialist, Wildlife Section, Illinois Natural History Survey; Sparks, 1980).

BENTHOS

The decline in use of the Illinois Valley by diving ducks and in the condition factor of commercially important species of fish is traceable to a decline in the benthos of the Illinois River. The question now becomes: what killed the small mollusks? Since fingernail clams go through an entire life cycle in 33 days (Gale, 1969), they are capable of quickly repopulating an area from which they have been eliminated. We hypothesized that there is some factor in the Illinois River which inhibits recolonization of the river from residual populations remaining in tributary streams.

The pattern of the die-off offers some clues to the source of the factor which affected the clams. A die-off was not recorded in the upper Illinois River, simply because the fingernail clam *(Musculium transversum)* either did not occur there because the habitat was

unsuitable or had already been eliminated by the time benthic surveys were initiated in the 1900's. Fingernail clams begin to appear in the ILlinois River again near the mouth of the Sangamon River. The source of the pollutant is probably not agricultural, because the Sangamon drains the fertile corn-growing region of central Illinois. The most likely explanation is that an upstream source introduces a toxic material which is gradually diluted or broken down as the water moves downstream.

PREVIOUS RESEARCH

Sparks, Sandusky, and Paparo (1981) found that certain layers of sediment in Quiver Lake, where fingernail clams had died out, were toxic to isolated clam gills. Two deeper layers of sediment were less toxic (depressed the particle transport rate less) than two shallower layers. Although the sediments were not dated, the findings are consistent with the fact that fingernail clams thrived in Quiver Lake until the die-off in 1955-58. If a toxic material were introduced to the river in 1955, it might be bound to the sediments deposited at that time (perhaps represented by the second layer). The deeper and older sediments (third and fourth layers) would not contain the toxicant. The decline in toxicity from the second layer to the surficial layer might reflect a reduction of toxicant input into the river or dilution by greater volumes of sediment eroded from farm lands and banks of tributary streams.

In order to determine whether Illinois River water still contains a factor toxic to fingernail clams, Sparks, Sandusky, and Paparo (1981) and Sparks and Sandusky (1981) exposed *Musculium transversum* from the Mississippi River to raw Illinois River water continuously pumped into a laboratory at Havana. Three bioassays were conducted at different times, and in two of the three mortality in the raw river water was significantly greater than in clean well water. Treatment of raw river water by filtration through sand, sand and clinoptilolite, or sand and carbon significantly improved survival.

The experiments with Quiver Lake sediments and raw Illinois River water demonstrated that both contain factors which are lethal to fingernail clams. At least one of the toxic factors is ammonia. Fingernail clam survival was enhanced in treatments which reduced un-ionized ammonia levels in raw Illinois River water (Sparks and Sandusky, 1981). Moreover, un-ionized ammonia levels in the Illinois River exceed concentrations which are lethal to fingernail clams exposed in the laboratory for six weeks (Sparks and Sandusky, 1981) and which cause inhibition of the cilia on clam gills (Anderson, Sparks, and Paparo, 1978).

Other factors, besides ammonia, may contribute to toxicity in the Illinois River. For example, the gills of fingernail clams are sensitive to heavy metals (Anderson, Sparks, and Paparo (1978), and sediments in the Illinois River contain higher concentrations of heavy metals than sediments in tributary streams (Mathis and Cummings, 1971). Fluoridation of public water supplies was increasing in the 1950's, when the die-off of fingernail clams occurred. Evanston, Illinois, was one of several cities in which fluoridation trials were initiated in 1945-1947 (Burgstahler, 1977). The city of Chicago started fluoridating its drinking water in August, 1956 (personal communication, 20 December, 1978, Roger Selburg, Division of Public Water Supplies, Illinois Environmental Protection Agency) and Peoria initiated fluoridation in July, 1958 (personal communication, 19 December, 1978, Hubert Hall, Supervisor, Peoria Water Works).

RESEARCH DESIGN

The original research design was to determine effects of fluoride, lead, cadmium, and sediments from two lakes along the Illinois River and from the Keokuk Pool, Mississippi River, on gill function, shell formation, survival, growth and reproduction of the fingernail clam. The tests which were completed are marked with an X in the table below:

Toxicant	Determine Effects of Toxicant on: survival, growth, reproduction	ciliary activity of gills	shell formation, composition
flouride	X	X	
lead	X	X	
cadmium		X	
sediments	X	X	

Shells of clams exposed to fluoride, lead and lake sediments have been saved and will be analyzed later, but the data were not available at the time this report was prepared. The effects of lead on the gills had been determined as part of a previous project, B-097-ILL (Anderson, Sparks, and Paparo, 1978), but the results are included in this report, for comparison with effects on clam growth and survival. The effects of cadmium on clam survival and growth were not determined.

The effects of sediments from four lakes, rather than three lakes, were determined. Turner Lake, a bottomland lake along the Illinois River near the Great Bend at Hennepin (Figure 5) was added to the list contained in the original proposal. Turner Lake is three miles upstream from Lake DePue and 92 miles upstream from Quiver Lake, near Havana. Lake DePue is a bottomland lake connected to the Illinois River, where a smelter once discharged its wastes. Quiver Lake is where Paloumpis and Starrett (1960) documented a die-off of fingernail clams. Sediments were collected from these three lakes, rather than from the main channel of the river, because the lakes serve as sediment traps, whereas the channel is scoured by current and dredged to maintain a 9-foot navigation channel. Sediment from the Keokuk Pool, Mississippi River, where fingernail clams are still abundant, served as a control.

If the relative toxicity of the Illinois River sediments increases in the upstream direction to Lake DePue, and then decreases at Turner Lake, Lake DePue and the smelter would be implicated as the source of toxic sediment. If the toxicity is greater in Turner Lake than in DePue, then the source probably lies further upstream.

Fluoride was selected for testing for reasons given above. The choice of the metals lead and cadmium was governed by results of an experiment in which fingernail clams were inadvertently exposed to water contaminated with the following concentrations of metals: 2.7 mg/l lead, 0.42 mg/l cadmium, 0.05 mg/l chromium, 0.09 mg/l cobalt, 0.52 mg/l copper, and 0.62 mg/l zinc (Anderson, Sparks and Paparo, 1978). Fingernail clams exposed to the contaminated water developed curved shells, and all the clams eventually died. Subsequent analysis of the shells by X-ray microprobe techniques revealed that the normal calcium-silicon ratio was reversed in the deformed regions of the shell. An additional reason for selecting these two heavy metals is that they occur at relatively high levels in the sediments in Lake DePue and Sawmill Lake (Lee and Stall, 1977) and a mainstem lake of the Illinois River, Peoria Lake (Mathis and Cummings, 1971). Sawmill Lake (near Henry), Lake DePue, and Peoria Lake are all in the reach of the river where the fingernail clam die-off occurred (Figure 5).

METHODS

COLLECTION OF FINGERNAIL CLAMS

Fingernail clams were collected from Keokuk Pool (Pool 19) of the Mississippi River using an 18-foot boat equipped with a crane and Ponar grab sampler. The samples were partially washed by pressure-sieving the bottom material through a 30-mesh screen with a 12-volt battery-operated water pump. The clams and a portion of sediment not washed through the screen were transported to the laboratory at Havana, Illinois, in 37-liter plastic coolers equipped with aerators and partially filled with Mississippi River water.

All clams collected on a particular date were regarded as a single "stock", and were maintained separately from stocks obtained on other dates. Each stock was assigned an identification number in order according to date.

MAINTENANCE OF FINGERNAIL CLAMS

Clams were kept in the transport cooler, with aeration, to acclimate them to the temperature of the laboratory. When the difference in water temperature between the cooler and the laboratory aquaria was less than 3°C, the clams were transferred. The aquaria received well water from a small diluter modified from the design of Mount and Brungs (1967).

The clams were fed a concentrated suspension of the green alga *Scenedesmus quadricauda*, which is abundant in Keokuk Pool. The alga was batch cultured in flasks on a staggered basis, so that a flask containing a maximum density of algal cells was available every 2-3 days. The algal suspension was delivered to the aquaria automatically every five minutes, by a system incorporated in the diluter.

Within a few days after being collected, duplicate groups of 20 clams were removed from the sediment in the stock aquaria and placed in 100-mm petri dishes. The dishes were covered with plastic snap-on lids

in which a 50-mm hole was cut to allow circulation of water. The holes were subsequently covered with 30-mesh nylon screen, after we discovered that small clams (2-4 mm in shell length) could climb the sides of the dish and along the underside of the plastic cover, and exit the hole. The petri dishes were placed back in the stock aquaria, and the growth and survival of the clams were measured after two and four weeks. The purpose of the monitoring was to prevent use of unhealthy or dormant stocks in the bioassays.

Growth was determined by measuring the maximum length of the shell to the nearest 0.1 mm with an ocular micrometer. When clams died, their valves gaped because the elastic hinge ligament was no longer opposed by the adductor muscles.

Benjamin and Burley (1978) have indicated that fingernail clams may be detritus feeders. The sediment from the Mississippi River which was brought back to the laboratory in the cooler was sieved through a 30-mesh screen to remove predators, such as leeches, and refrigerated at 2°C. The sediment was replaced with refrigerated sediment, warmed to room temperature, when the growth and survival of the clams was checked every two weeks.

Gale (1972) suggested that the decomposing remains of dead clams inhibit the growth of live clams kept in the same container. Dead clams or shells were removed from the petri dishes when they were checked every two weeks.

COLLECTION AND PREPARATION OF SEDIMENT SAMPLES

Sediment cores were taken from the mid-point of each of the three lakes along the Illinois River. Sediment samples from the Keokuk Pool, Mississippi River, were taken upstream from Montrose, Iowa, in water approximately one meter deep near River Mile 375.4. Duplicate cores were extruded from the 10-cm diameter steel corer onto a plastic tray. The top five cm of both cores were thoroughly mixed together, and an aliquot placed in a jar and shipped to Southern Illinois

University for testing on clam gills. Another aliquot was taken to the Natural History Survey Laboratory for testing on intact clams.

At Southern Illinois University, the sediment was stored overnight in a refrigerator, warmed to room temperature the next day and used immediately. The wet mud was added to one liter of invertebrate physiological solution, until the desired sediment concentration (in mg/liter) was obtained. Sediment concentrations were determined by passing a measured volume of the test solution through a membrane filter, then weighing the air-dried sample. The average particle size of the sample was measured under a microscope with an ocular micrometer and particle counts were made with a hemocytometer. Concentrations of the test solutions made from the various lake sediments were adjusted until they were nearly equal (Table 1). All concentrations were well below the levels which affect clam gills by direct mechanical action or clogging (Anderson et al, 1978).

TABLE 1

Physical Characteristics of Sediment Suspensions Tested on Fingernail Clam Gills

Values are means ± standard deviations

Site	Date Sampled	Particle Size (μm)	Density (mg/l)	Concentration (Particles/L)
Keokuk Pool	8 Nov. 1980	14.4±3.1	71.2±10.4	$(5.3\pm0.8)\times10^5$
Quiver Lake	18 Nov. 1980	13.3±4.6	57.4±6.2	$(4.6\pm0.7)\times10^5$
Lake DePue	28 Nov. 1980	15.6±4.6	69.7±9.2	$(5.1\pm1.0)\times10^5$
Turner Lake	4 Dec. 1980	12.9±2.5	44.3±3.8	$(4.8\pm0.9)\times10^5$
Keokuk/Turner 1:1 Mix		13.7±4.3	61.2±6.2	$(5.1\pm0.7)\times10^5$
Keokuk/Turner 1:2 Mix		14.1±1.9	57.2±2.2	$(5.9\pm1.1)\times10^5$

Intact clams were exposed to the sediments from the four sites by placing the clams in petri dishes containing the sediments, and renewing the sediments every two weeks with refrigerated sediment which had been warmed to room temperature.

BIOASSAYS USING CLAM GILLS

Fingernail clams were acclimated at least one week in invertebrate physiological solution in Instant Ocean Aquaria at a temperature of 17°C and a pH ranging from 7.8 to 8.2. Gills were excised from the clams and placed in petri dishes, where a continuous flow of standard physiological solution or solution to which sediments or toxicants had been added was maintained by means of metering pumps. Temperatures and dissolved oxygen concentrations in the petri dishes were monitored by thermistor meters and membrane electrodes.

Two responses were measured in the sediment bioassays: Particle transport rates and beating rates of the lateral cilia. Particle transport rates (in mm/sec) were calculated from the time it took for particles to move across the gill a known distance within the microscope field. Ciliary beating rates were measured using two coupled microscopes, with a stroboscopic light serving as the substage light source for both microscopes (Anderson et al. 1978). The rate of ciliary beating (in beats per sec) was measured by manually synchronizing the rate of flashing of the light with the rate of beating of the lateral cilia, which beat in a metachronal pattern. Synchronization was achieved when the metachronal wave appeared to stand still. The beating rate was shown on a digital display. Some sediments and toxicants caused very low ciliary beating rates, which could not be measured by the above apparatus. In these cases, we used a shutter mechanism and a high-intensity light source, similar to a movie projector, to accurately measure low ciliary beating rates. Average transport rates or beating in a microscope field showing approximately 50 gill filaments were determined. Gills from 5-7 clams

were observed and the means and standard deviations are reported in the results. Other responses were recorded, such as complete inhibition of ciliary beating or a shift from the normal metachronal pattern to a synchronous or erratic pattern.

Ciliary beating rates only were measured in bioassays in which gills were exposed to toxic cations or anions because the solutions contained no particles which could be used for determining particle transport rates. The following reagent grade salts were tested: cadmium chloride ($CdCl_2$), lead nitrate ($Pb\ [NO_3]_2$), sodium fluoride (NaF) and a salt containing two toxic ions, cadmium fluoride (CdF_2). Sodium, chloride, and nitrate show little or no toxicity to fingernail clams (Anderson et al. 1978). Cadmium fluoride was tested to determine whether the toxic effects of cadmium and fluoride are additive. Concentrations in the results section are reported in terms of the toxic ion, in mg/l.

BIOASSAYS USING INTACT CLAMS

The test chambers were 37.8-liter glass aquaria with outlets to maintain the volume at 23 liters. The test chambers were immersed in a water bath to control water temperature. Water flow from a diluter (Mount and Brungs 1967) was approximately 100 ml/min and was checked three times a day. The diluter delivered a logarithmic series of toxicant concentrations via flow splitters, so that two aquaria were maintained at each concentration. An automatic feeding system (separate from the one used on the stock aquaria) delivered 100 ml of algal suspension to each test chamber every 5 minutes. Algal concentrations were measured by procedures in Standard Methods (American Public Health Association, 1980).

Each test chamber held two petri dishes initially containing 20 clams each (for a total of 40 clams exposed to each test solution). Sieved sediment collected from the Mississippi River on the same date the clams were collected was added to the petri dishes at the beginning of the bioassay and at two-week intervals thereafter, when survival and growth were checked. The sediment was refrigerated when collected and warmed to room temperature before it was added to the petri dishes.

Fingernail clams are very active and climb the sides of glass containers, so the petri dishes were covered with a plastic snap-on lid in which a 50-mm hole was cut to allow circulation of water. The hole was covered with 30-mesh nylon screen. Because the clams were small, active, fragile-shelled, and nearly transparent, some were unavoidably lost when the contents of the petri dishes were being sieved at two-week intervals. If any clams were missing at each two-week check, the numbers missing were recorded and are reported in the tables of results. Of course, missing clams were not counted as survivors or dead clams. Dead clams were easily identified under the microscope, because the shells gaped and were usually empty. When a clam dies, the elastic hinge ligament forces the shell open and the soft body parts decay and disappear within a few hours.

Water temperatures, dissolved oxygen concentrations and pH in the test chambers were measured two or three times per week, and alkalinity once a week. Concentrations of fluoride, cadmium and lead in the gill bioassays were calculated from the amount of salt added to the test solution. In the bioassays with intact clams, samples for lead analysis were taken three times during the bioassay. The samples were filtered through a 0.45 micron membrane filter, and the filtrate analyzed for soluble lead. The filter was then treated with one ml of concentrated nitric acid. A few ml of ultrapure water was added and

the solution decanted. The filter was washed with ultrapure water and the solution and wash combined, made up to 50 ml and analyzed for lead. The latter results were considered to represent the precipitated or insoluble lead. Samples were analyzed on an atomic absorption spectrophotometer. Fluoride was measured according to Standard Methods (American Public Health Association, 1980).

Variance tests (Snedecor and Cochran 1967) were used to determine whether there were significant differences in clam mortality between treatments. Mortalities in duplicate petri dishes within each test chamber were pooled. An analysis of variance, ANOVA (Steel and Torrie 1960), was used to determine whether there were significant differences in shell lengths of clams exposed to the different test solutions and the clean well water. If the mortality in a test solution was significantly different from mortality in the well water control, the length data were not analyzed, because of the possibility that mortality was size-dependent. If, for example, the rate of uptake and effect of a toxic substance depended on the body volume or gill surface area of a clam, mortality would be size-dependent, and size differences between clams exposed to the various treatments and the well water would reflect differential mortality rather than differential growth. All differences were considered significant at a probability, $P \leq .05$.

RESULTS

RESPONSE OF CLAMS TO CADMIUM

Response of Gills

Cadmium concentrations greater than .030 reduced ciliary beating rates below that of controls (Table 2 and Figure 7). A concentration of .060 mg/l reduced the ciliary beating rate to approximately 50% of the normal value (EC50). Concentrations greater than .095 mg/l caused complete ciliary arrest.

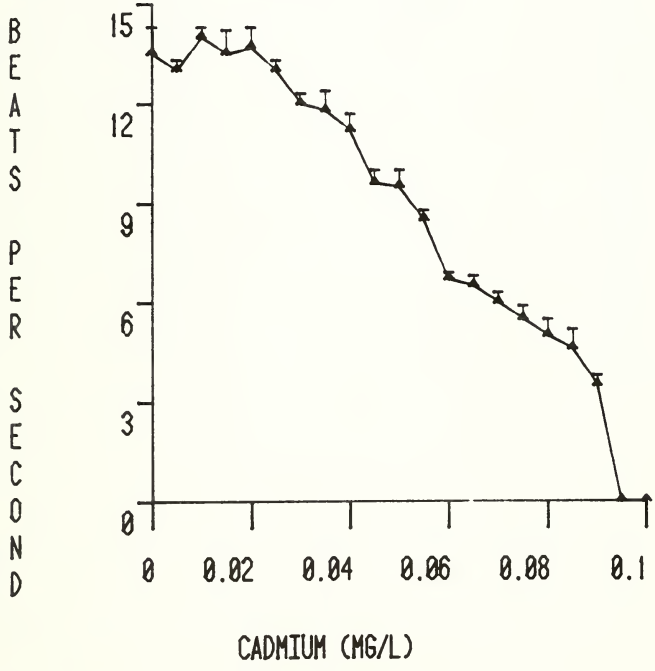

Figure 7. Effects of cadmium on ciliary beating rate of fingernail clam gills. Exposure time at each concentration = 10 minutes. Values are means of 5-7 gills, with the positive standard deviation in brackets.

TABLE 2

Response of Fingernail Clam Gills to Cadmium, Added as Cadmium Chloride, $CdCl_2$ (Exposure time = 10 minutes)

Concentration Added Cd^{++} (mg/l)	Average Ciliary Beating Rate ± S.D. (Beats/sec.)	Maximum Ciliary Beating Rate	% of Maximum Rate with no Cd
.000	13.5 ± 1.0	14.5	100.0
.005	13.0 ± 0.5	13.5	93.1
.010	14.0 ± 0.5	14.5	100.0
.015	13.5 ± 0.9	14.4	99.3
.020	13.7 ± 0.8	14.5	100.0
.025	13.0 ± 0.5	13.5	93.1
.030	12.0 ± 0.5	12.5	86.2
.035	11.8 ± 0.8	12.6	86.9
.040	11.2 ± 0.7	11.9	82.1
.045	9.6 ± 0.6	10.2	70.3
.050	9.5 ± 0.7	10.2	70.3
.055	8.5 ± 0.5	9.0	62.1
.060	6.7 ± 0.4	7.1	49.0
.065	6.5 ± 0.5	7.0	48.3
.070	6.0 ± 0.5	6.5	44.8
.075	5.5 ± 0.6	6.1	42.1
.080	5.0 ± 0.7	5.7	39.3
.085	4.6 ± 0.8	5.4	37.2
.090	3.5 ± 0.5	4.0	27.6
.095	0	0	0
.100	0	0	0

RESPONSE OF CLAMS TO LEAD

Response of Gills

Anderson et al. (1978) reported that the ciliary beating rates of gills from small clams, approximately the same size used in the present research, were reduced to 50% of the normal level by lead concentrations between 0.002 and 0.020 mg/l. We used the value of 0.020 as a conservative estimate of the EC50.

Response of Intact Clams

The water chemistry, temperature and algal concentrations maintained in the test chambers during the lead bioassays are given in Tables 3 and 4. The mean values for dissolved lead concentrations in the test chambers increase in a logarithmic series as expected, based on the amount of lead nitrate added to the diluter. There is slight overlap of the ranges in the three higher concentrations. However, values for particulate and total lead do not follow the expected pattern, and the ranges are wide and overlap considerably. We observed that the diluter and test chambers accumulated a precipitate, presumably insoluble lead salts formed in the water. The precipitate may have been dislodged and sporadically flushed from the diluter cells and it is likewise possible that the precipitate was stirred up from the bottom when the water samples were dipped from the test chambers. Because of these sampling problems, the soluble lead concentrations reported in Table 3 probably best represent the relative differences in lead concentrations to which the fingernail clams were exposed.

The soluble and insoluble forms of other heavy metals, such zinc, are known to differ in their toxicity to fish (Cairns et al. 1971). Our bioassay was not designed to determine whether soluble and insoluble forms of lead differ in their toxicity to fingernail clams. We recommend that bioassays be conducted with lead salts differing in solubility to answer this question.

TABLE 3

Water Chemistry and Temperature During Lead Bioassay
Ranges are given below means (N = 3)

Lead (mg/l)			Temp. (°C)	pH	Dissolved Oxygen (mg/l)	Total Alkalinity (mg/l as $CaCO_3$)	Hardness (mg/l as $CaCO_3$)
Soluble	Particulate	Total					
<.018[a]	<.019	<.037	20.2	7.79	9.35	293	349
<.016-.022	<.016-.026	<.032-.048	19.5-21.2	7.74-7.95	8.89-9.71	292-294	341-354
.035	.188	.223	20.3	7.82	9.26	292	358
.030-.038	.059-.265	.095-.303	19.7-21.2	7.78-7.88	8.80-9.68	---	341-374
.052	.334	.386	20.3	7.84	9.23	292	358
.038-.072	.130-.447	.176-.496	19.7-21.2	7.80-7.89	8.79-9.60	---	341-374
.087	.442	.529	20.4	7.82	9.22	295	357
.053-.119	.083-.669	.171-.788	19.8-21.3	7.79-7.87	8.79-9.60	294-296	341-372
.189	.192	.381	20.4	7.82	9.14	291	357
.095-.271	.043-.199	.294-.537	19.8-21.3	7.75-7.86	8.78-9.59	290-292	341-372
.503	.146	.649	20.3	7.81	9.23	291	356
.266-.654	.038-.244	.421-.898	19.8-21.2	7.77-7.87	8.77-9.62	290-292	341-372

[a] Well water control, no added lead. Detection limit = 0.016 mg/l.

TABLE 4

Algae Concentrations (colonies per ml) Delivered to Test Chambers During Lead Bioassay.
(100ml delivered to each chamber every 5 minutes)
Ranges are given below means

WEEK					
1	2	3	4	5	6
1,498	1,903	2,048	2,054	1,995	1,945
1,196-1,808	1.674-2,129	1,797-2,205	1,925-2,205	1,925-2,077	1,896-2,018

There were no significant differences in survival of fingernail clams exposed to soluble lead concentrations ranging from .018 to .503 mg/l (the highest concentration tested) for up to six weeks (Table 5).

There were significant differences in the mean shell lengths of clams exposed to the various lead concentrations. In the table below, concentrations in which mean shell lengths were not significantly different are connected by a line:

Soluble Lead (mg/l)	Length of Exposure			
	Initial	2 Weeks	4 Weeks	6 Weeks
<.018				
.035				
.052				
.087				
.189				
.503				

The shell lengths of the clams did not differ at the beginning of the bioassay, because the clams were purposely selected for uniformity. After two weeks, clams exposed to .018 and .035 mg/l lead differed significantly in shell length from a group which included the four higher concentrations. After four weeks, there were three groups with significantly different shell lengths, but all three groups showed some overlaps. At the end of six weeks however, shell lengths in the two lowest concentrations differed significantly from lengths in the four higher concentrations. If we assume that the soluble lead is the form that is toxic to fingernail clams, then the lowest concentration which produces a significant effect on clam growth after six weeks of exposure lies between .035 and .052 mg/l. The equivalent total lead concentrations (Table 3) are .223 and .386 mg/l.

TABLE 5

Results of Lead Bioassay

Soluble Lead (mg/l)	Number Alive	Number Missing	Mean Shell Length (mm)	Variance	Standard Deviation	Range (mm)	Mean Growth Increment (mm)
INITIAL							
<.018[a]	40	--	2.06	0.00	0.07	1.9-2.2	--
.035	40	--	2.06	0.01	0.07	1.9-2.2	--
.052	40	--	2.06	0.00	0.06	1.9-2.2	--
.087	40	--	2.06	0.00	0.07	1.9-2.2	--
.189	40	--	2.06	0.00	0.07	1.9-2.2	--
.503	40	--	2.03	0.00	0.06	1.9-2.1	--
2 WEEKS							
<.018	37	1	2.22	0.01	0.12	2.0-2.4	0.16
.035	37	--	2.18	0.01	0.10	2.0-2.4	0.12
.052	38	--	2.14	0.01	0.09	2.0-2.4	0.08
.087	37	--	2.14	0.01	0.08	2.0-2.3	0.08
.189	39	--	2.14	0.01	0.09	1.9-2.3	0.08
.503	37	1	2.11	0.01	0.09	1.9-2.3	0.08
4 WEEKS							
<.018	37	--	2.25	0.01	0.11	2.1-2.4	0.03
.035	37	--	2.20	0.01	0.11	2.1-2.4	0.02
.052	36	--	2.16	0.01	0.10	2.0-2.4	0.02
.087	36	--	2.14	0.01	0.08	2.0-2.3	--
.189	38	--	2.15	0.01	0.09	1.9-2.3	0.01
.503	36	--	2.12	0.01	0.09	2.0-2.3	0.01
6 WEEKS							
<.018	37	--	2.25	0.01	0.12	2.1-2.5	--
.035	37	--	2.22	0.01	0.10	2.1-2.4	0.02
.052	35	--	2.16	0.01	0.10	2.0-2.4	--
.087	36	--	2.15	0.01	0.08	2.0-2.3	0.01
.189	38	--	2.17	0.01	0.10	1.9-2.3	0.02
.503	33	--	2.15	0.01	0.09	2.0-2.4	0.03

[a]Well water control, no added lead.

Lead Levels in the Illinois River

In order to determine whether lead concentrations in the Illinois River might be in the range which affects fingernail clams, the total lead concentrations reported by the U.S. Geological Survey (1981 and 1982) are compared to the total lead concentration which appeared to produce a significant decrease in shell growth of fingernail clams in our experiments (Figures 8 and 9). The midpoint of the range within which a significant decline in clam shell lengths occurred is .304 mg/l total lead, and is shown as a horizontal line in Figures 8 and 9. The U.S. Geological Survey reported soluble, insoluble and total lead concentrations, at Marseilles and Valley City, but only total lead at the other six sampling stations along the Illinois River in 1979-1980. In 1980-1981, soluble and total lead were measured at all eight stations. Values shown in Figures 8 and 9 are total lead. Grab samples were taken once a month or less frequently, so it is impossible to know how long fingernail clams in the Illinois River were exposed to the concentrations shown in Figures 8 and 9. Nevertheless, the data can be used to show whether lead concentrations in the Illinois River ever approach levels which might affect fingernail clams.

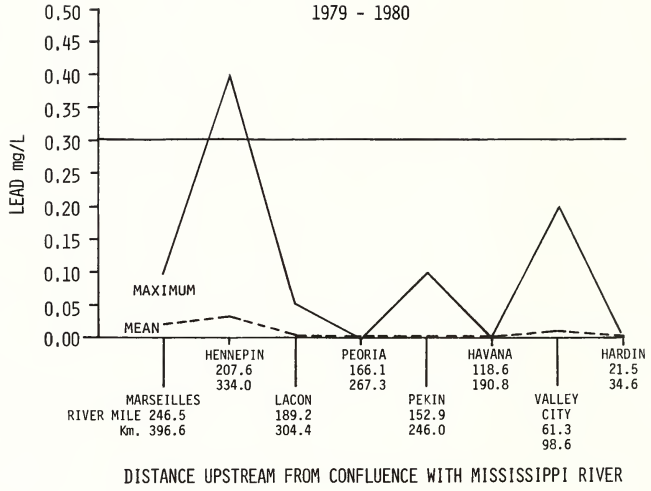

Figure 8. Mean and maximum total lead concentrations at eight sampling stations in the Illinois River 1 October 1979 - 30 September 1980. Source: U.S. Geological Survey, 1981. Horizontal line = concentration which reduced growth of fingernail clams in the laboratory.

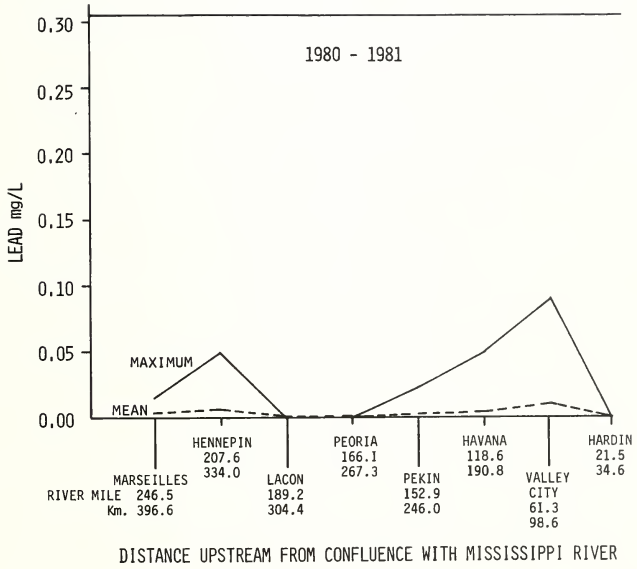

Figure 9. Mean and maximum total lead concentrations at eight sampling stations in the Illinois River, 1 October 1980 - 30 September 1981. Source: U.S. Geological Survey, 1982. Horizontal line = concentration which reduced growth of fingernail clams in the laboratory.

The mean total lead concentrations in the Illinois River between 1 October 1979 and 30 September 1980 were well below .304 mg/l, but the maximum concentration at Hennepin (River Mile 207.6) exceeded this value (Figure 8). Between 1 October 1980 and 30 September 1981 both the mean and maximum total lead concentrations were well below .304 mg/l (Figure 9).

We reiterate the importance of determining the toxicity of the soluble and particulate forms of lead to fingernail clams. If it is the soluble form of lead which is toxic, our results show that a soluble lead concentration between .035 and .052 mg/l (Table 3) adversely affects the growth of fingernail clams. Based on the samples taken at Valley City (downstream) and Marseilles (upstream), more than half the total lead usually occurs in the insoluble or particulate form. The maximum soluble lead concentration reported for the Illinois River between 1 October 1979 and 30 September 1981 was 0.005 mg/l, considerably below the levels which affected clams in our bioassays.

RESPONSE OF CLAMS TO FLUORIDE

Response of Gills

A fluoride concentration of 0.75 mg/l reduced the ciliary beating rate of fingernail clam gills by approximately 50% (Table 6 and Figure 10). A fluoride concentration of 1 mg/l caused ciliary arrest (Table 6 and Figure 10).

Response of Intact Clams

Water chemistry, temperature, and algal concentrations delivered to the test chambers are reported in Tables 7 and 8. The well water contained 0.12 mg/l fluoride. The ranges of concentrations in the test solutions were fairly narrow and did not overlap (Table 7). Approximately 2×10^5 algal colonies were delivered to each test chamber every 5 minutes during the eight-week bioassay (Table 8).

TABLE 6

Response of Fingernail Clam Gills to Fluoride Added as Sodium Fluoride, NaF. (Exposure time = 10 minutes)

Concentration Added F^-(mg/l)	Average Ciliary Beating Rate ± S.D. (Beats/sec.)	Maximum Ciliary Beating Rate	% of Maximum Rate with no F^-
.00	12.5 ± 0.7	13.2	100.0
.05	12.0 ± 0.4	12.4	93.9
.10	11.5 ± 0.6	12.1	91.7
.15	11.0 ± 0.3	11.3	85.6
.20	10.5 ± 0.7	11.2	84.9
.25	10.5 ± 0.4	10.9	82.6
.30	9.5 ± 0.3	9.8	74.2
.35	9.0 ± 0.9	9.9	75.0
.40	8.5 ± 0.7	9.2	69.7
.45	8.5 ± 0.4	8.9	67.4
.50	8.0 ± 0.8	8.8	66.7
.55	7.5 ± 0.3	7.8	59.1
.60	7.5 ± 0.5	8.0	60.6
.65	7.0 ± 0.5	7.5	56.8
.70	7.0 ± 0.2	7.2	54.6
.75	6.0 ± 0.5	6.5	49.2
.80	5.0 ± 0.6	5.6	42.4
.85	4.5 ± 0.5	5.0	37.9
.90	3.0 ± 0.4	3.4	25.8
.95	1.5 ± 0.8	2.3	17.4
1.00	0	0	0

TABLE 7

Water Chemistry and Temperature During Fluoride Bioassay
Ranges are given below means.

Fluoride (mg/l)	Temp. (°C)	pH	Dissolved Oxygen (mg/l)	Total Alkalinity (mg/l as $CaCO_3$)
0.12[a]	20.9	7.80	8.79	228.4
0.10-0.13	19.2-21.8	7.75-7.88	8.17-9.15	212-248
0.54	20.9	7.82	8.73	229.6
0.52-0.55	19.1-21.8	7.76-7.89	7.82-9.15	216-250
0.85	20.9	7.82	8.72	231.2
0.80-0.92	19.1-21.7	7.77-7.89	7.9-9.13	216-250
1.80	21.0	7.81	8.75	231.6
1.65-1.96	19.1-21.7	7.77-7.88	8.28-9.14	220-252
2.82	20.9	7.81	8.73	232.4
2.44-3.45	19.1-21.6	7.77-7.89	8.1-9.09	220-252
4.56	20.9	7.83	8.73	234.8
4.25-4.75	19.0-21.7	7.79-7.92	8.21-9.16	222-256

[a]Well water control, no added fluoride.

TABLE 8

Algae Concentrations (colonies per ml) Delivered to Test Chambers
During Fluoride Bioassay.
(100ml delivered to each chamber every 5 minutes)
Ranges are given below means

Week							
1	2	3	4	5	6	7	8
1929	2369	2220	1928	2280	1845	1690	1574
1680-2187	2129-2625	1849-2444	1791-2100	2135-2502	1680-2106	1522-1820	1388-1773

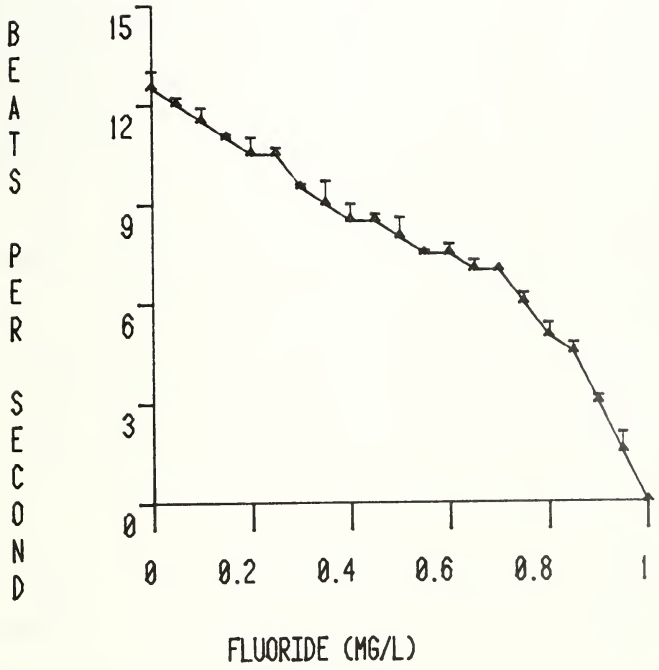

Figure 10. Effects of fluoride on ciliary beating rate of fingernail clam gills. Exposure time at each concentration = 10 minutes. Values are means of 5-7 gills, with positive standard deviations in brackets.

There were no significant differences in clam mortality between the test concentrations after exposures of two weeks and six weeks. After four weeks, mortalities in clams exposed to 0.54 and 2.82 mg/l fluoride differed significantly from control clams exposed to well water containing no added fluoride (Table 9). After eight weeks, mortalities in 2.82 mg/l fluoride significantly exceeded mortalities in the controls, while mortalities in all the other concentrations did not differ significantly from the controls (Table 9). These results are displayed graphically in Figure 11, which shows that the 95% confidence limits for mortality in the 2.82 mg/l concentration do not overlap the confidence limits around the well water control.

Clams grew in all test concentrations, but the greatest increase in mean shell length occurred in the well water control (Table 9). In the table below, mean shell lengths did not differ significantly in concentrations falling along the vertical lines. The only significant differences in shell lengths occurred after two weeks and six weeks of exposure. For example, the table below shows that after two weeks shell lengths of clams exposed to 0.12, 0.54, 1.80, 2.82, and 4.56 did not differ significantly from each other, but did differ from the group comprised of shell lengths in the 0.54, 0.85, 2.82, and 4.56 concentrations.

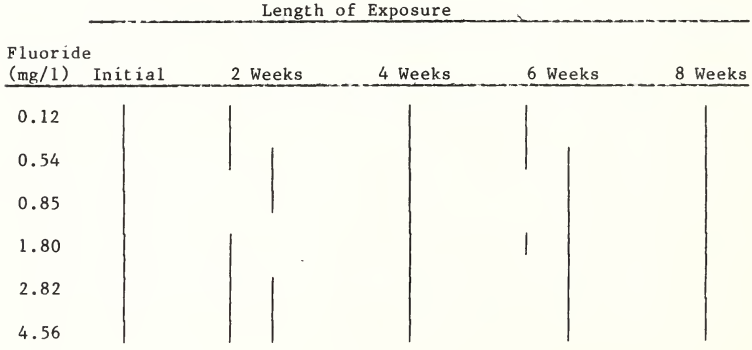

TABLE 9

Results of Fluoride Bioassay

	Number Alive	Number Missing	Mean Shell Length (mm)	Variance	Standard Deviation	Range (mm)	Mean Growth Increment (mm)
INITIAL							
0.12[a]	40	--	2.18	0.02	0.16	1.9-2.5	--
0.54[b]	40	--	2.17	0.03	0.16	1.9-2.5	--
0.85[b]	40	--	2.16	0.02	0.16	1.8-2.5	--
1.80[b]	40	--	2.15	0.02	0.14	1.8-2.4	--
2.82[b]	40	--	2.16	0.02	0.14	1.8-2.5	--
4.56[b]	40	--	2.16	0.04	0.20	1.8-2.5	--
2 WEEKS							
0.12	40	--	2.31	0.03	0.18	2.0-2.7	0.13
0.54	36	--	2.28	0.02	0.16	2.0-2.6	0.11
0.85	36	--	2.19	0.02	0.02	1.9-2.4	0.03
1.80	39	--	2.30	0.02	0.16	1.9-2.6	0.15
2.82	33	2	2.25	0.03	0.16	2.0-2.6	0.09
4.56	37	--	2.25	0.04	0.19	2.0-2.7	0.09
4 WEEKS							
0.12	40[c]	--	2.33	0.17	0.41	2.1-3.4	0.02
0.54	32	--	2.30	0.02	0.15	2.0-2.6	0.02
0.85	35	--	2.21	0.02	0.15	1.9-2.5	0.02
1.80	35	--	2.29	0.02	0.15	1.9-2.6	--
2.82	32	--	2.27	0.03	0.16	2.0-2.6	0.02
4.56	36	--	2.29	0.04	0.19	2.0-2.7	0.04
6 WEEKS							
0.12	35	1	2.39	0.06	0.25	2.1-3.4	0.06
0.54	30	--	2.32	0.02	0.16	2.1-2.6	0.02
0.85	31	1	2.22	0.03	0.16	1.9-2.5	0.01
1.80	30	1	2.31	0.03	0.16	1.9-2.6	0.02
2.82	26	--	2.25	0.02	0.15	2.0-2.6	0.02
4.56	31	--	2.26	0.04	0.20	2.0-2.7	--
8 WEEKS							
0.12	30	--	2.38	0.03	0.18	2.1-2.8	--
0.54	26	--	2.33	0.03	0.17	2.1-2.7	0.01
0.85	22	--	2.26	0.02	0.14	2.0-2.5	0.04
1.80	19	--	2.32	0.02	0.15	2.1-2.6	0.01
2.82	16	--	2.28	0.02	0.16	2.1-2.6	0.03
4.56	18	1	2.32	0.04	0.21	2.1-2.7	0.06

[a]Well water control, no added fluoride.
[b]Mean concentrations in test chambers, as fluoride, F^-, mg/l, based on 3 samples taken during bioassay.
[c]One 1.7mm clam, born in the control chamber, was removed and not counted as part of the control population.

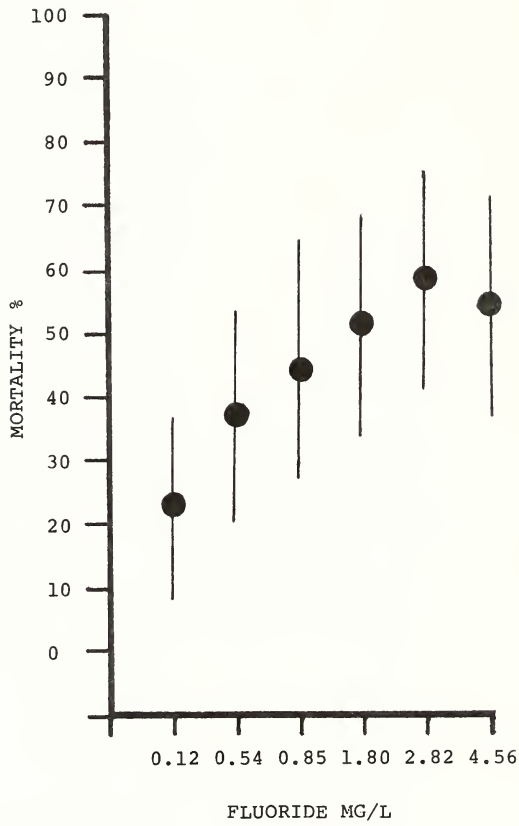

Figure 11. Mortality of fingernail clams exposed to six fluoride concentrations for eight weeks. The lowest concentration (0.12 mg/l) was in well water to which no fluoride was added. Brackets denote 95% confidence limits.

The above table shows no consistent differences in mean shell lengths in the various fluoride concentrations throughout the bioassay.

Some clams developed curved shell margins or cracked shells and others showed an abnormally slow response to the substage illumination when they were examined under the microscope at two week intervals. By the end of eight weeks, 6 out of 132 shells were deformed. One of these shells occurred in the well water control, and the others occurred in the test solutions. At each two-week interval, approximately 20% of the clams exposed to test solutions containing added fluoride crawled away from the microscope illumination very slowly, as though they were partially sedated. Most of these clams were in the low fluoride concentrations, rather than in the two highest concentrations. We know of no known mechanism for fluoride toxicity which would explain the deformed shells or retarded movements.

Fluoride Levels in the Illinois River

Dissolved fluoride concentrations are measured up to 12 times a year at Marseilles (River Mile 246.5) and Valley City (River Mile 61.3). Between 1 October 1980 and 30 September 1981 fluoride concentrations were usually higher at Marseilles than Valley City, probably because of the discharge of fluoridated municipal water from the Chicago area. The maximum fluoride concentration at Marseilles was 0.7 mg/l, which is considerably below the concentration of 2.82 which significantly increased the mortality of fingernail clams in our experiment, following eight weeks of exposure. It is close to the concentration of 0.75 mg/l which caused a 50% reduction in the ciliary beating rate of clam ;gills, but the intact clams apparently have a defensive mechanism which is not available to the isolated gills.

RESPONSE OF CLAMS TO MIXTURES OF CADMIUM AND FLUORIDE

Response of Gills to Mixtures

Table 10 and Figure 12 shows the response of clam gills to mixtures of cadmium and fluoride produced by adding the salt cadmium fluoride (CdF_2) to physiological saline. The concentration of cadmium in the mixture which produced a 50% reduction in ciliary beating rate in 10 minutes (10-min EC50) was .046 mg/l and the fluoride concentration was approximately .023 mg/l. The EC50's determined for test solutions containing only one toxicant were .06 mg/l for cadmium and .75 mg/l for fluoride. Hence the cadmium concentration in the mixture contained approximately .77 of the EC50 for cadmium (.046 ÷ .060) and .03 of the EC50 for flouride (.023 ÷ .750). If the toxic contributions from cadmium and fluoride in the mixture are added, .77 + .03 = .80, the mixture is predicted to have .80 of a 10-min EC50. Since the mixture was more toxic than predicted, it is evident that cadmium and fluoride are synergistic, i.e. their effects are more than additive. The significance of these findings is that combinations of toxicants in the Illinois River may be more toxic to fingernail clams than would be predicted from results of bioassays using single toxicants.

RESPONSE OF CLAMS TO SEDIMENTS

Response of Gills

Effects of Sediments on Ciliary Beating Rates. Isolated gills from fingernail clams were exposed to sediments from the four lakes for sixty minutes. Ciliary beating rates in sediment from Keokuk Pool increased slightly during the test; sediments from Lake DePue caused a slight, but significant decline in beating rates; and sediments from both Quiver Lake and Turner Lake depressed ciliary beating rates slightly as soon as they were added to the test chambers, followed by an increase after 10-20 minutes, and an abrupt decline after 20-30 minutes (Table 11 and Figure 13).

TABLE 10

Response of Fingernail Clam Gills to a Mixture of Fluoride and Cadmium, Added as Cadmium Fluoride, CdF_2
(Exposure time = 10 minutes)

Concentration Added			Average Ciliary Beating Rate ± S.D. (Beats/sec.)	Maximum Ciliary Beating Rate	% of Maximum Rate with no F^-
CdF_2 (mg/l)	Cd^{++} (mg/l)	F^- (mg/l)			
.0000	.0000		13.0 ± 1.3	14.3	100.0
.0050	.0037	.0013	13.2 ± 0.6	13.8	96.5
.0100	.0075	.0025	12.9 ± 0.9	13.8	96.5
.0150	.0112	.0038	13.0 ± 0.8	13.8	96.5
.0200	.0150	.0050	13.0 ± 1.1	14.1	98.6
.0250	.0187	.0063	12.3 ± 0.7	13.0	90.9
.0300	.0224	.0076	11.7 ± 1.1	12.8	89.5
.0350	.0262	.0088	11.0 ± 0.5	11.5	80.4
.0400	.0299	.0101	10.3 ± 0.6	10.9	76.2
.0450	.0336	.0114	9.3 ± 0.7	10.0	69.3
.0500	.0374	.0126	7.9 ± 0.9	8.8	61.5
.0550	.0411	.0139	7.3 ± 0.7	8.0	55.9
.0600	.0448	.0152	6.1 ± 0.4	6.5	45.5
.0650	.0486	.0164	5.5 ± 0.8	6.3	44.1
.0700	.0523	.0177	5.6 ± 0.6	6.2	43.4
.0750	.0561	.0189	5.0 ± 0.3	5.3	37.1
.0800	.0598	.0202	4.5 ± 0.5	5.0	35.0
.0850	.0635	.0215	3.5 ± 0.6	4.1	28.7
.0900	.0673	.0227	1.0 ± 0.4	1.4	9.8
.0950	.0710	.0240	0	0	0
.1000	.0747	.0253	0	0	0

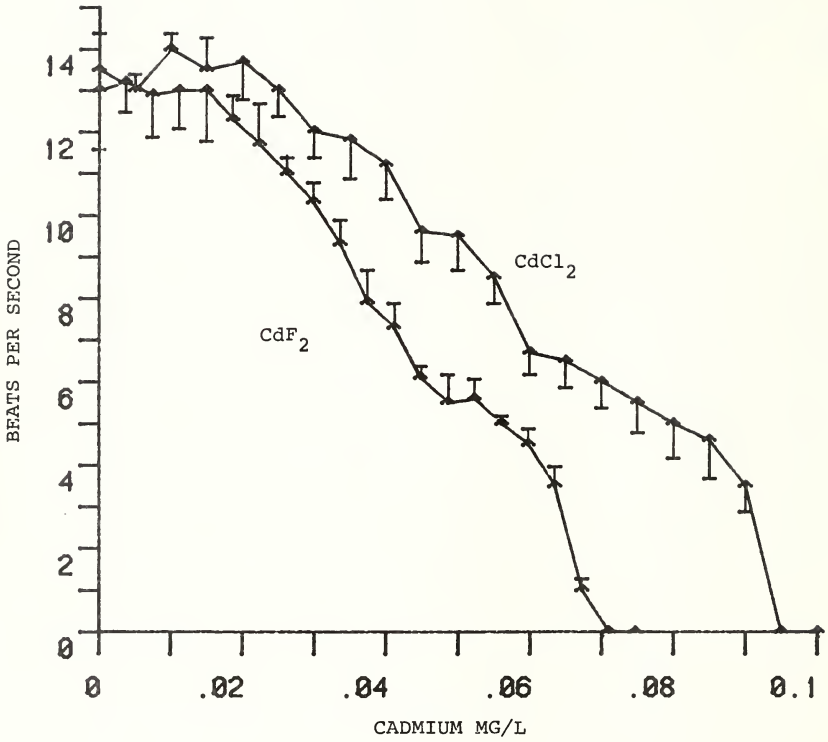

Figure 12. Effects of cadmium fluoride and cadmium chloride on ciliary beating rate of fingernail clam gills.

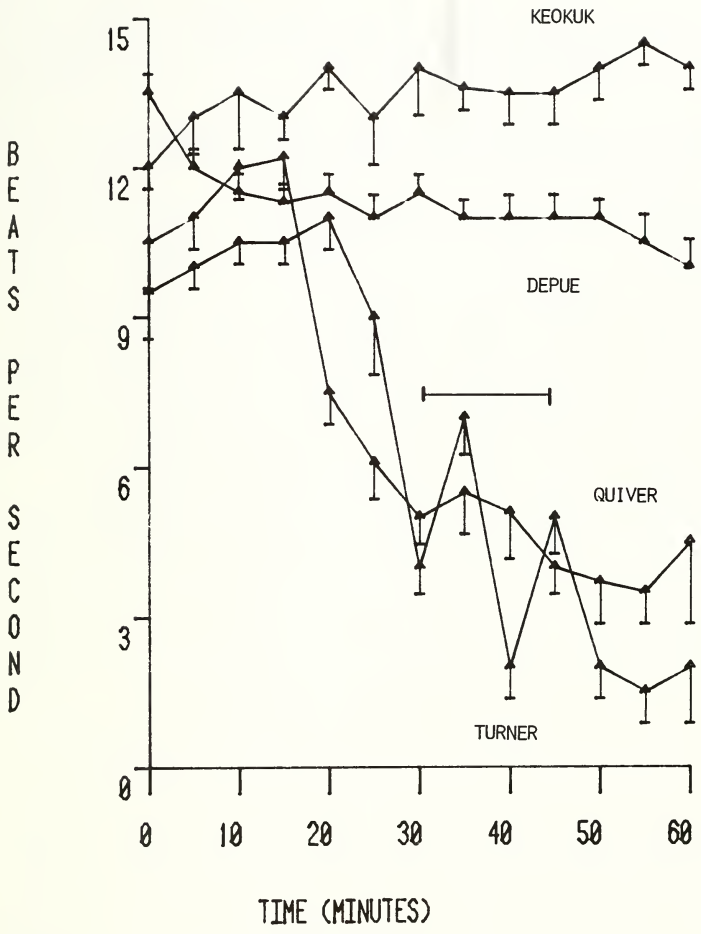

Figure 13. Effects of sediments on ciliary beating rates.

TABLE 11

Response of Fingernail Clam Gills to Sediments from Keokuk Pool, Mississippi River and Three Lateral Lakes of the Illinois River

Values are average beating rates (beats per second) of lateral cilia on clam gills ± the standard deviations. Test conditions: pH=7.4±0.2, O_2=8.0ppm±0.6, Temperature=205°C±0.5.

	MISSISSIPPI	ILLINOIS RIVER		
TIME	KEOKUK	QUIVER	DE PUE	TURNER
0	12.0 ± 0.3	10.5 ± 0.8	13.5 ± 0.5	9.5 ± 0.8
5	13.0 ± 0.6	11.0 ± 0.5	12.0 ± 0.5	10.0 ± 0.3
10	13.5 ± 1.0	12.0 ± 0.5	11.5 ± 0.5	10.5 ± 0.3
15	13.0 ± 0.3	12.2 ± 0.5	11.3 ± 0.5	10.5 ± 0.3
20	14.0 ± 0.3	7.5 ± 0.5	11.5 ± 0.5	11.0 ± 0.5
25	13.0 ± 0.8	6.1 ± 0.6	11.0 ± 0.6	9.0 ± 1.0
30	14.0 ± 0.8	5.0 ± 0.4	11.5 ± 0.5	4.0[a] ± 0.4
35	13.6 ± 0.3	5.5 ± 0.7	11.0 ± 0.5	7.0[a] ± 0.6
40	13.5 ± 0.5	5.1 ± 0.8	11.0 ± 0.6	2.0[a] ± 0.5
45	13.5 ± 0.5	4.0 ± 0.4	11.0 ± 0.6	5.0[a] ± 0.6
50	14.0 ± 0.5	3.7 ± 0.7	11.0 ± 0.5	2.0[a] ± 0.5
55	14.5 ± 0.3	3.5 ± 0.5	10.5 ± 0.7	1.5 ± 0.5
60	14.0 ± 0.3	4.5 ± 0.5	10.0 ± 0.7	2.0 ± 1.0

[a] abnormal, synchronous beating

Sediments from Turner Lake appeared to be the most toxic, because they produced the lowest ciliary beating rates and abnormal, synchronous beating (Table 11 and Figure 13).

Effects of Sediments on Particle Transport Rates. Particle transport rates on clam gills exposed to sediment from Keokuk Pool, Mississippi River, increased for approximately 35 minutes, then appeared to stablize at approximately 0.50 mm/sec (Table 12 and Figure 14). The degree of toxicity in the sediments from the three lakes along the Illinois River followed the same pattern as for the ciliary beating rates: Lake DePue least toxic, Quiver Lake next, and Turner most toxic (Table 12 and Figure 14). Unlike the ciliary beating rates, the particle transport rates were not significantly depressed until 20-30 minutes of exposure (Figure 14).

Effects of Sediment Mixtures on Gills. Nontoxic sediments from Keokuk Pool was mixed with the most toxic lake sediment from the Illinois River in ratios of 1:1 and 1:2 (Keokuk:Turner). The decline in particle transport rates (Table 13 and Figure 15) and ciliary beating rates (Table 14 and Figure 16) was proportional to the amount of Turner Lake sediment. Abnormal, synchronous beating was only observed with the 1:2 mixture (Table 14).

Response of Intact Clams

There were no significant differences in growth rate or mortality of fingernail clams exposed to sediments from the four sites for six weeks (Table 15). However, the mean shell lengths (2.52 mm) and survival (74%) of clams in Keokuk sediment was greater than in sediments from the Illinois River (Table 15). The smallest shell length (2.43 mm) and lowest survival (27%) occurred in sediment from Lake DePue (Table 15).

TABLE 12

Response of Fingernail Clam Gills to Sediments from the Keokuk Pool, Mississippi River and Three Lateral Lakes of the Illinois River

Values are average particle transport rates (mm per second) ± the standard deviations. Test conditions: pH = 7.4 ± 0.2, O_2 = 8.0 ppm ± 0.6, temperature = 20.5°C ± 0.5.

TIME (min)	MISSISSIPPI KEOKUK	ILLINOIS RIVER QUIVER	ILLINOIS RIVER DE PUE	ILLINOIS RIVER TURNER
0	.22 ± .02	.28 ± .04	.33 ± .04	.26 ± .03
5	.27 ± .02	.32 ± .03	.38 ± .02	.30 ± .01
10	.38 ± .02	.34 ± .02	.42 ± .02	.32 ± .02
15	.44 ± .02	.37 ± .03	.47 ± .02	.34 ± .04
20	.46 ± .01	.36 ± .03	.48 ± .02	.33 ± .03
25	.49 ± .02	.37 ± .01	.48 ± .02	.30 ± .02
30	.51 ± .02	.34 ± .03	.48 ± .02	.24 ± .02
35	.53 ± .01	.35 ± .03	.46 ± .01	.20 ± .02
40	.51 ± .02	.33 ± .03	.44 ± .01	.14 ± .02
45	.52 ± .01	.31 ± .02	.42 ± .02	.12 ± .02
50	.50 ± .03	.31 ± .02	.44 ± .03	.10 ± .01
55	.52 ± .02	.26 ± .02	.42 ± .02	.08 ± .01
60	.48 ± .02	.24 ± .02	.42 ± .02	.06 ± .02

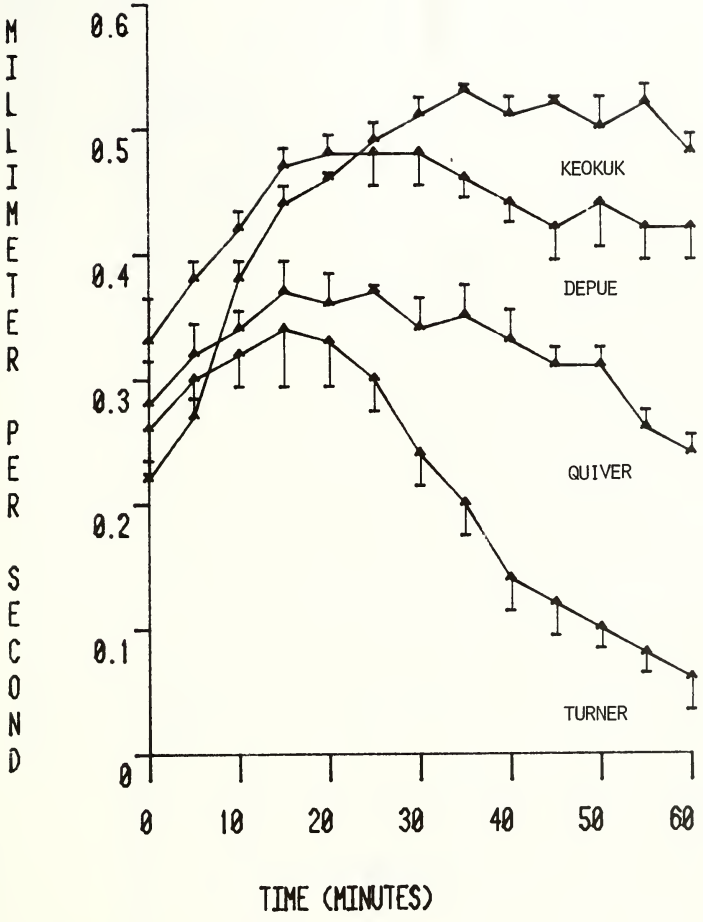

Figure 14. Effects of sediments on particle transport rates.

TABLE 13

Effect of Mixed Sediments on Particle Transport Rates (mm per second) of Clam Gills

Values are means ± standard deviations

TIME (hrs)	KEOKUK	KEOKUK:TURNER 1:1 Mix	KEOKUK:TURNER 1:2 Mix
0.00	.32 ± .04	.32 ± .04	.32 ± .04
0.25	.36 ± .02	.36 ± .02	.36 ± .02
0.50	.39 ± .03	.39 ± .03	.39 ± .03
0.75	.38 ± .02	.38 ± .02	.38 ± .02
1.00	.38 ± .01	.38 ± .01	.38 ± .01
1.25	.38 ± .02	.34 ± .02	.28 ± .02
1.50	.37 ± .02	.33 ± .03	.20 ± .02
1.75	.38 ± .01	.32 ± .03	.12 ± .03
2.00	.36 ± .03	.28 ± .02	.06 ± .02
2.25	.35 ± .02	.24 ± .04	.04[a] ± .01
2.50	.33 ± .03	.20 ± .02	.02[a] ± .02
2.75	.32 ± .02	.14 ± .02	.03[a] ± .02
3.00	.31 ± .04	.12 ± .02	.02 ± .02

[a] abnormal, synchronous beating of cilia on gills

TABLE 14

Effect of Mixed Sediments on Ciliary Beating Rates (beats per second) of Clam Gills

Values are means ± standard deviations

TIME (hrs)	KEOKUK	KEOKUK:TURNER 1:1 Mix	KEOKUK:TURNER 1:2 Mix
0.00	13.5 ± 0.6	13.5 ± 0.6	13.5 ± 0.6
0.33	14.0 ± 0.5	14.0 ± 0.5	14.0 ± 0.5
0.67	13.5 ± 0.5	13.5 ± 0.5	13.5 ± 0.5
1.00	14.4 ± 0.6	14.4 ± 0.6	14.4 ± 0.6
1.33	14.0 ± 0.6	12.5 ± 0.7	12.0 ± 0.8
1.67	14.3 ± 0.4	12.5 ± 0.8	10.0 ± 1.2
2.00	13.5 ± 0.3	12.0 ± 1.0	3.6[a] ± 0.9
2.33	14.0 ± 0.4	11.3 ± 0.8	6.5[a] ± 0.6
2.67	13.2 ± 0.4	10.7 ± 1.0	4.5[a] ± 0.6
3.00	13.0 ± 0.6	10.3 ± 0.8	5.0 ± 0.6

[a] abnormal synchronous beating of cilia on gills

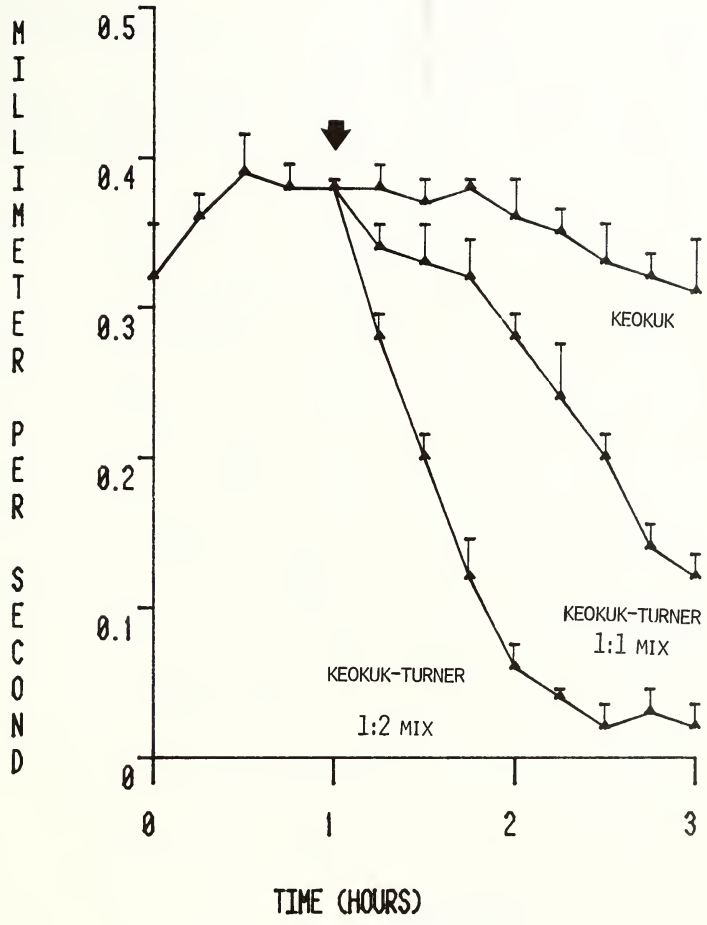

Figure 15. Effects of sediment mixtures on particle transport rates.

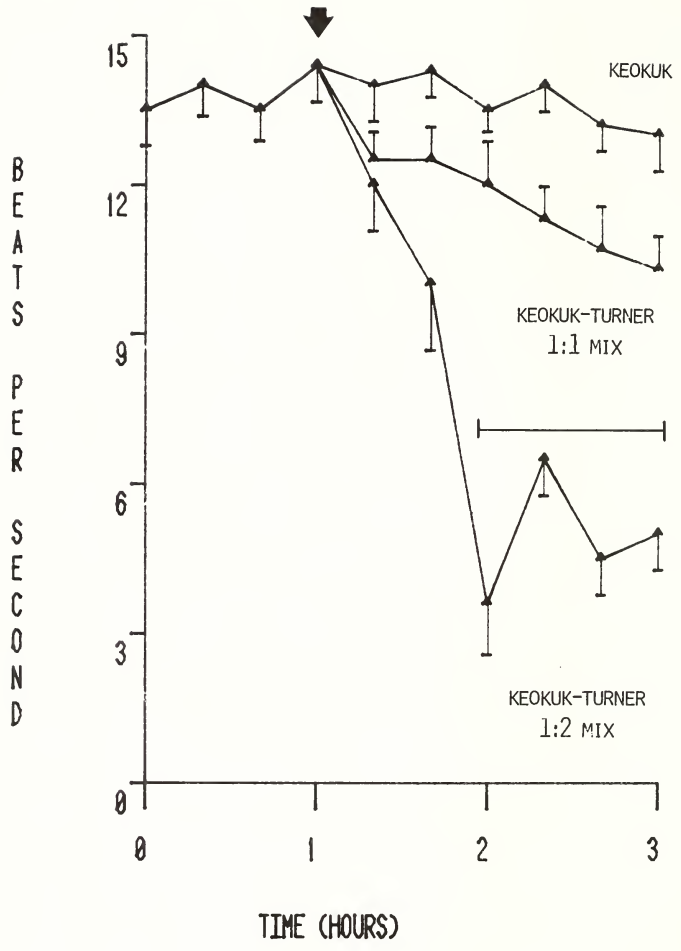

Figure 16. Effects of sediment mixtures on ciliary beating rates.

TABLE 15

Results of Sediment Bioassay

Source of Sediment	Number Alive	Number Missing Missing	Mean Shell Length (mm)	Variance	Standard Deviation	Range (mm)	Mean Growth Increment (mm)
Initial							
Keokuk	40	--	2.45	0.02	0.15	2.1-2.7	--
Quiver	40	--	2.43	0.02	0.15	2.1-2.7	--
Depue	40	--	2.44	0.03	0.17	2.1-2.7	--
Turner	40	--	2.44	0.03	0.16	2.2-2.8	--
2 Weeks							
Keokuk	33	1	2.50	0.02	0.14	2.2-2.7	0.05
Quiver	37	0	2.47	0.02	0.14	2.2-2.7	0.04
Depue	34	1	2.46	0.03	0.18	2.1-2.8	0.02
Turner	34	0	2.45	0.03	0.17	2.2-2.8	0.01
4 Weeks							
Keokuk	32	1	2.49	0.02	0.15	2.2-2.8	--
Quiver	30	0	2.48	0.02	0.14	2.2-2.7	0.01
Depue	28	2	2.43	0.03	0.18	2.1-2.8	--
Turner	31	0	2.45	0.03	0.16	2.2-2.8	0.01
6 Weeks							
Keokuk	29	1	2.52	0.02	0.14	2.2-2.8	0.03
Quiver	25	0	2.50	0.02	0.16	2.2-2.7	0.02
Depue	18	2	2.43	0.04	0.19	2.2-2.9	--
Turner	27	0	2.44	0.03	0.19	2.1-2.8	--

DISCUSSION

The table below summarizes results obtained using both the gill response (ciliary beating rate) and survival of intact clams. Gill responses are expressed as percentages of the normal ciliary beating rates, prior to addition of the lake sediments. Survival rates are also expressed as percentages. Survival of clams exposed to sediments from lakes along the Illinois River was poorer after six weeks of exposure than for clams exposed to sediment from the Mississippi River, although the differences were not significantly significant. The same sediments tested on fingernail clam gills produced statistically significant decreases in ciliary beating rates.

	Miss. R.	Illinois River		
	Keokuk Pool	Quiver Lake	Lake DePue	Turner Lake
Gill Response	100%	33%	74%	21%
Survival	72%	62%	47%	68%

Particle transport rates on the gills also showed significant declines, in the same pattern as the beating rates.

The table below compares the concentrations which produced 50% declines in ciliary beating rates or significant declines in the growth or survival of intact clams with the maximum concentrations reported in the Illinois River between 1 October 1979 and 30 September 1981 (U.S. Geological Survey 1981 and 1982). The effects of cadmium were tested on clam gills only, not on intact clams.

	Concentration (mg/l)		
	Fluoride	Lead	Cadmium
Gill response	0.75	0.02	0.06
Growth or mortality	2.82	0.30	----
Illinois River	0.80	0.40	0.01

The maximum fluoride concentration was well below the level which affected the growth or mortality of intact clams. The maximum lead concentration slightly exceeded the total lead concentration which significantly reduced the growth of fingernail clams in a six-week exposure. Since the grab samples for lead analysis were taken from eight sites on the Illinois River twelve times a year or less, it is impossible to determine how long clams in the river were exposed to the maximum concentration. Because this high concentration occurred only once in two years, the clams were probably not exposed as long as six weeks. Total cadmium concentrations in the river were below levels which affected clam gills.

Sparks and Sandusky (1981) and Sparks et al. (1981) demonstrated that un-ionized ammonia levels in the Illinois River sporadically exceed levels which affected the growth and survival of fingernail clams in the laboratory. Moreover, they showed that treatment of Illinois River water to remove ammonia and sediment dramatically improved the survival of the clams. It appears that un-ionized ammonia and an unidentified toxic material in sediments are the major factors preventing recolonization of the Illinois River by fingernail clams. Sporadic peaks in fluoride, lead and cadmium in water may contribute to the toxicity exerted by ammonia and the sediment factor (especially if the peaks happen to coincide), but are probably not the major toxic factors.

RELATION OF THIS RESEARCH TO WATER RESOURCES PROBLEMS

This project approach and results can be used by regulatory and management agencies interested in restoration of degraded rivers. The results have drawn attention to the fact that sediment quality, as well as water quality can be limiting to aquatic life.

PUBLICATIONS RESULTING FROM THIS RESEARCH

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